Conclusion: Low dose of MI substantially improved LV relaxation in association with stimulation of Ca^{2+} ATPase activity in sarcoplasmic reticulum

1121-35 Total Body Endothelin-1 Kinetics in Normal Subjects and Heart Failure Patients: A New Radiotracer Approach

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Endothelin-1 (ET-1) is elevated in CHF, although its kinetics are poorly understood in humans. We describe a technique of ^{125}I-labelled ET-1 (^{125}I-ET-1) infusion to determine total body ET-1 kinetics.

Methods/Results: 5 normals and 5 CHF patients were studied. Over 5 minutes, 50 microcuries of ^{125}I-ET-1 were infused. Hemodynamics and blood samples were obtained 31 times over 240 minutes. After C_{18} column extraction, plasma was counted in a gamma counter. ET-1 plasma levels were measured using an enzyme immunoassay technique. Kinetics of ^{125}I-ET-1 clearance were modelled through standard computer modelling systems. Baseline circulating ET-1 levels were elevated in the CHF patients (2.82 ± 1.06 pg/ml vs 0.97 ± 0.43, P < 0.05). There were no changes in hemodynamics or circulating ET-1 levels with ^{125}I-ET-1 infusion in either group. The kinetics of ET-1 clearance were best described by a three compartment model with a prolonged terminal half life (455 ± 59 minutes, normals; 20689 ± 20172, CHF). The steady state volume of distribution was large but significantly smaller in CHF patients (34.9 ± 9.9 L/kg vs 23.7 ± 6.4 P < 0.05). The clearance rate of ET-1 from plasma was slower in CHF patients (0.066 ± 0.0088 L/kg/min vs 0.036 ± 0.012, P = 0.05).

Conclusions: This study describes a three compartment pharmacokinetic profile for ET-1. Its terminal half life and plasma volume of distribution are significantly larger than previous reports. The pharmacokinetic profile of CHF patients is altered and may explain its elevated circulating levels.

1121-36 The Neuronal Norepinephrine Transporter in Experimental Heart Failure After Supracoronary Aortic Banding in the Rat

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Background: Cardiac net secretion of norepinephrine (NE) is increased in congestive heart failure. To further elucidate the role of the neuronal norepinephrine transporter (NET, uptake$_1$) experiments were performed in rats with supracoronary aortic banding.

Methods and Results: Compared to sham operated control rats, rats with aortic banding were characterized by myocardial hypertrophy (heart weight 1.77 ± 0.08 vs. 1.22 ± 0.02 g), pulmonary congestion (7.24 ± 0.38 vs. 4.09 ± 0.16 g), and increased plasma NE (4.4 ± 0.8 vs. 3.0 ± 0.6 nmol/l). In isolated perfused hearts functional impairment of NET was indicated by (i) the uptake of [^{3}H]-NE being reduced (20 ± 1 vs. 30 ± 2%) and (ii) pharmacological blockade of NET with desipramine (DMI, 300 nmol/l) leading to a diminished increase in endogenous NE release evoked by electrical field stimulation (4 Hz, 1 min) (S2/S1 1.2 ± 0.2 vs. 1.7 ± 0.1; S1: control; S2: after DMI) (n ≥ 6, p < 0.05 each). RT-PCR was used to study NET gene expression. In situ hybridization with the DIG-labeled PCR product revealed a dense hybridization in the perikarya of tyrosine hydroxylase positive cell bodies within the left stellate ganglion but neither in efferent cardiac nerves nor the heart. NET mRNA concentrations of the left stellate ganglion were quantified by competitive RT-PCR, as was 18S rRNA to control for intact RNA. However, there was no significant difference between aortic banded and sham operated rats (1.65 ± 0.11 vs. 1.81 ± 0.07 NET mRNA/18S rRNA × 10^{-4}, n = 6).

Conclusion: Cardiac NE uptake by the NET is impaired in aortic banded rats with heart failure despite unaltered NET gene expression.

1121-37 Endothelin Receptor Blockade Improves Left Ventricular Pressure-Volume Relationships Without Altering the Hypertrophic Response During Congestive Heart Failure in Rats

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Background: The role played by endothelin-1 (ET-1) in the progression of congestive heart failure (CHF) remains unknown. We investigated the effects of ET receptor blockade on cardiac dimensions and dynamics, and on gene expression markers of hypertrophy during CHF.

Methods: After ligation of the left coronary artery, rats were randomized to oral treatment with a non-selective ET receptor antagonist (Bosentan, 100 mg/kg/day, n = 11) or saline (n = 13) for 15 days starting 24 hours after the induction of myocardial infarction. Ten sham-operated rats received no treatment.

Results: Right ventricular wt./body wt. ratios were increased by 65% in both the CHF groups (p < 0.05). Echocardiography revealed an increase in left ventricular end-diastolic dimensions (LV EDD) from 5.7 ± 0.1 in the sham group to 8.7 ± 0.2 mm in the CHF-saline group, while fractional shortening (FS) was decreased from 32 ± 2 to 12 ± 1% (p < 0.05). Intervention with bosentan caused 35% reduction in LV dilatation (LVEDD 7.6 ± 0.3 mm) and 49% increase in FS (to 18 ± 1%) compared to vehicle (p < 0.05). Bosentan also reduced LV end-diastolic pressure from 23 ± 1 to 20 ± 2 mmHg and LV systolic pressure from 107 ± 3 to 99 ± 2 mmHg (p < 0.05). Expression of preproET-1, ANP, BNP, β-MHC and skeletal α-actin mRNAs were all markedly and significantly increased in non-ischemic myocardium in CHF. No differences were observed between the CHF groups.

Conclusion: Intervention with Bosentan dramatically improves LV pressure-volume relationships and cardiac dynamics without altering the hypertrophic response during CHF in rats.

1121-38 What Is the Source of Increased Myocardial Tumor Necrosis Factor-alpha in Heart Failure?

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Background: Tumor necrosis factor-alpha (TNF-a) is a pleotropic proinflammatory cytokine and plasma TNF-a level appears to cofer prognosis in patients with heart failure (CHF). However, there is debate regarding the source of the increased TNF-a. Some investigators argue that TNF-a is produced by immune activation at end stage CHF. We therefore studied TNF-a in a dog model of pacing-induced CHF in which there is little tissue inflammatory response.

Methods: Six dogs were right ventricular-paced at 250 beats/min for 3 weeks to severe CHF. Serial plasma levels (mean ± SE) of TNF-a (by ELISA, pg/ml) and norepinephrine (NE, by HPLC, pg/ml) are shown.

*p < 0.05 versus baseline

	Baseline	Week 1	Week 2	Week 3
TNF-a	8 ± 4	79 ± 27*	89 ± 25*	247 ± 98*
NE	237 ± 34	391 ± 62	445 ± 74*	644 ± 164*

Results: Evolving CHF is accompanied by a progressive increase in plasma TNF-a and NE. Immunohistochemical studies of the LV of the paced dogs revealed intense staining for TNF-a in the myocytes, endothelial and smooth muscle layers, but not in the fibroblasts. These changes were not observed in control normal dogs. Staining of the spleens from the paced dogs were similar to the controls.

Conclusion: Increased plasma and myocardial TNF-a in a model of CHF with little immune pathogenesis suggests a primary production of TNF-a in the heart without a requirement of immune activation.

1121-39 Ca^{2+} Release From Sarcoplasmic Reticulum Is Slowed Down in Heart Failure

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Background: In heart failure, little information is available as to the Ca^{2+} release function of SR which plays a major role on cardiac contractility.

Method: The SR vesicles were isolated from dog LV muscles (normal [N], n = 7; pacing induced heart failure [HF], n = 6). The time course of SR Ca^{2+} release was continuously monitored by a stopped-flow apparatus using arsenazoIII as Ca^{2+} indicator, and [^{3}H]ryanodine (Ry) binding assay was done using filtration method.

Result: As compared with N, LV end-diastolic pressure was elevated by 380 ± 91% (p < 0.01). The peak + dP/dt was decreased by 24 ± 13% (p < 0.01) while time constant of LV pressure decay during isovolumic relaxation was increased by 19 ± 8% (p < 0.01), indicating HF. The amount